



MOHANLAL  
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UDAIPUR



DEPARTMENT  
OF  
ZOOLOGY

# Topic

# Estimation of Haemoglobin

*B.Sc. 3<sup>rd</sup>*

*Practical of Zoology*

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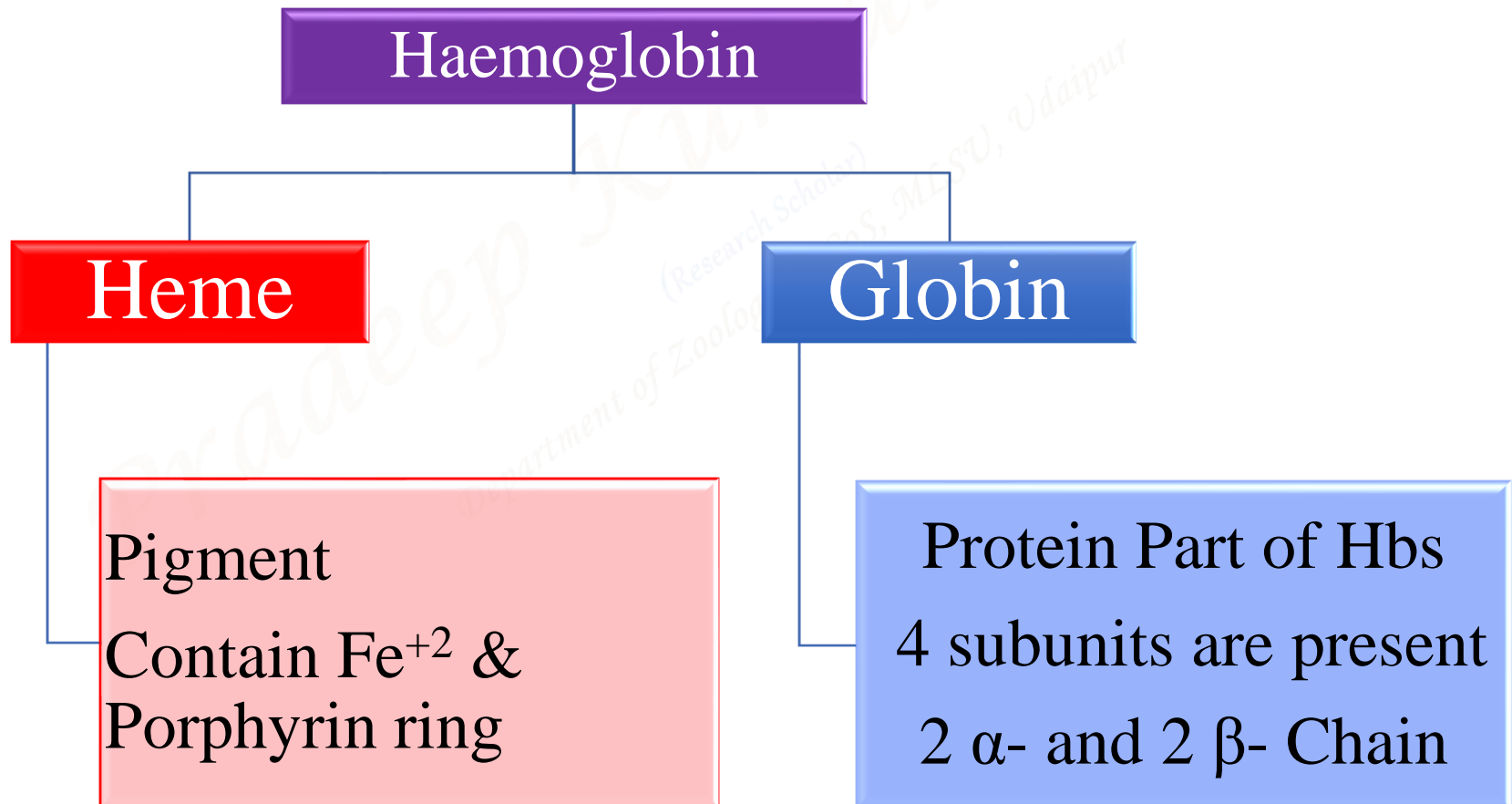
*University College of Science*

*Mohanlal Sukhadia University, Udaipur (Raj.)*

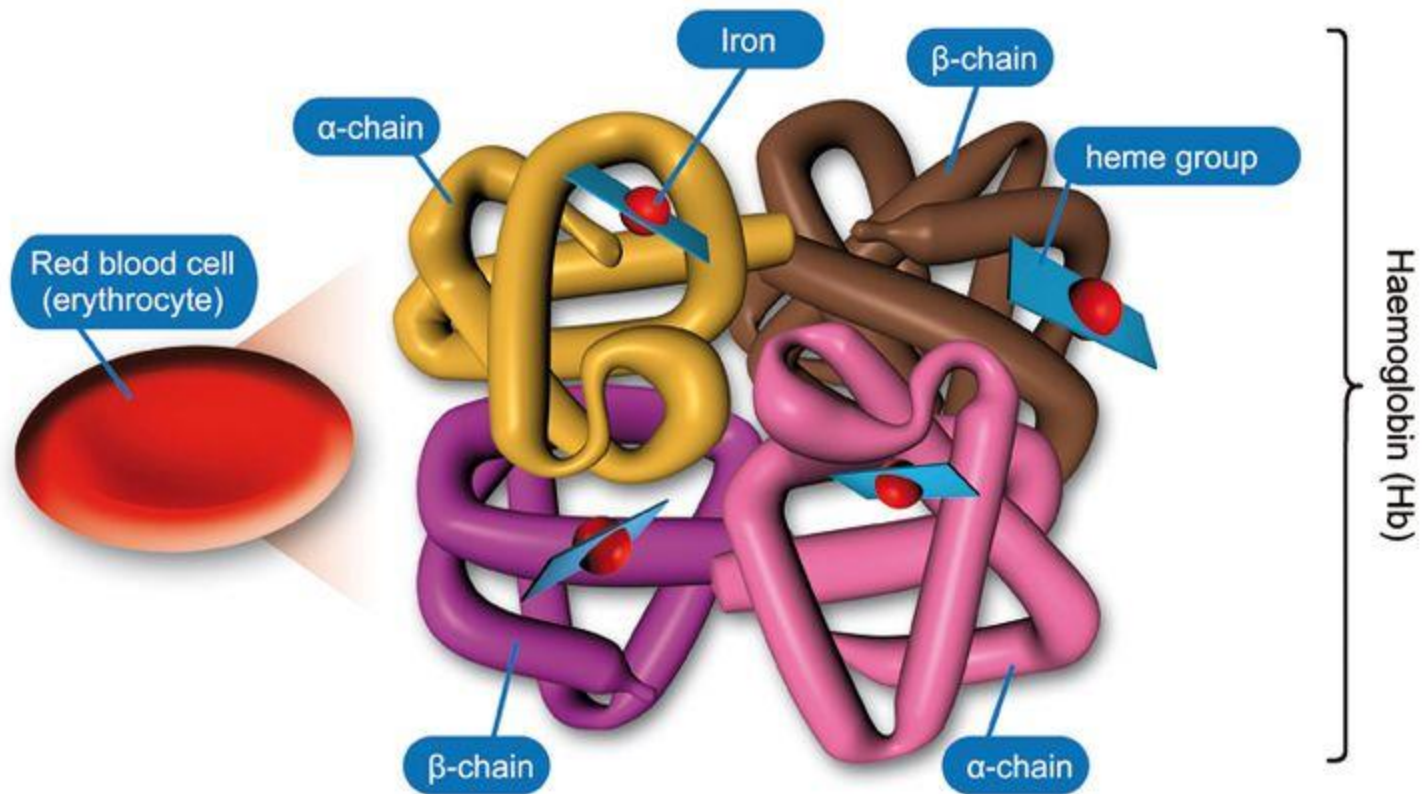
# Haemoglobin:

Hemoglobin is the protein molecule in red blood cells that carries oxygen from the lungs to the body's tissues and returns carbon dioxide from the tissues back to the lungs.

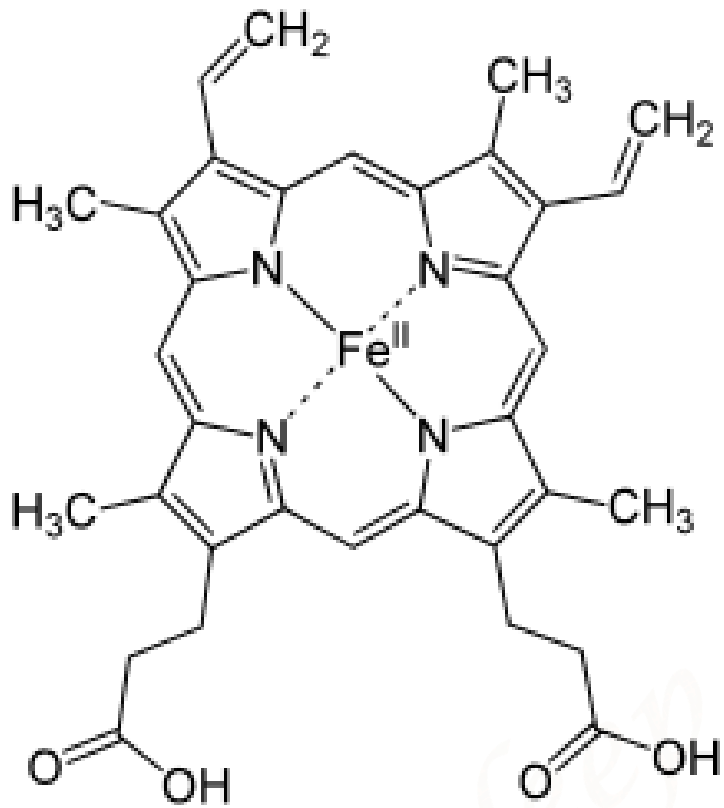
It is composed of four subunits, each with a **Heme** group and a **Globin** chain



## Structure of haemoglobin

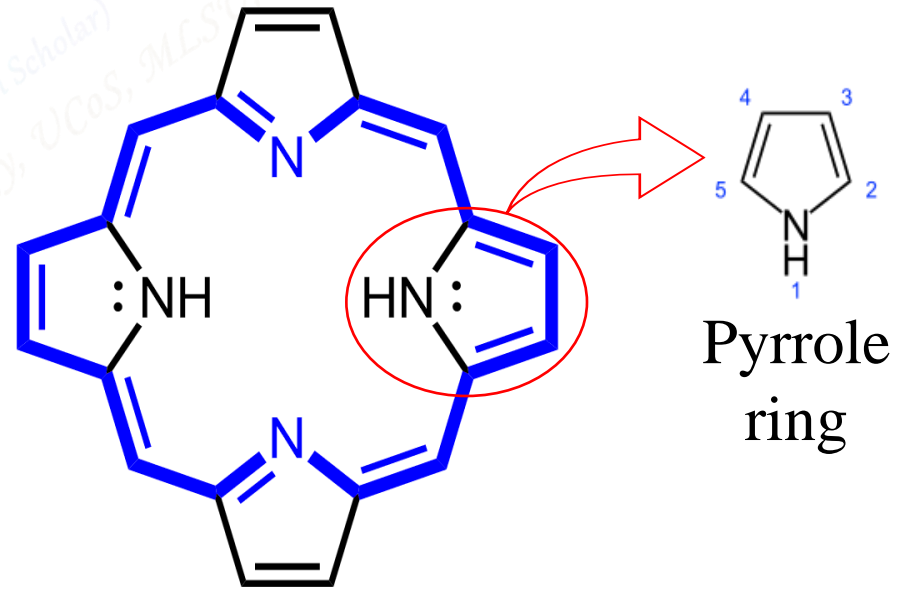


Each erythrocyte (RBC) contains ~270 million haemoglobin molecules



Haem pigment

Porphyrin ring + Fe ion



Porphyrin ring

Pyrrole ring

## Normal Values:

Male: 14 to 17 grams per deciliter (g/dL) or 140 to 170 gms per liter (g/L)

Female: 12 to 15 g/dL or 120 to 150 g/L

Female have approximately 12% lower than male.

The hemoglobin molecule has four binding sites for oxygen molecules: the iron atoms in the four heme groups

each Hb tetramer can bind four oxygen molecules.

1 g of Hb can combine with 1.39 ml of oxygen and in 100 ml of blood, there is about 15 g of Hb, so that 100 ml of blood has the capacity to bind ~20 ml of oxygen.

# Fluctuation in Hbs Amount:

## Low Amount:

Anemia caused by red blood cells dying earlier than normal (hemolytic anemia)

Bleeding from digestive tract or bladder, heavy menstrual periods

Kidney disease

Bone marrow being unable to produce new red blood cells. This may be due to leukemia, other cancers, drug toxicity, radiation therapy, infection, or bone marrow disorders

Poor nutrition (including low level of iron, folate, vitamin B12, or vitamin B6)

Low level of iron, folate, vitamin B12, or vitamin B6

## High Amount:

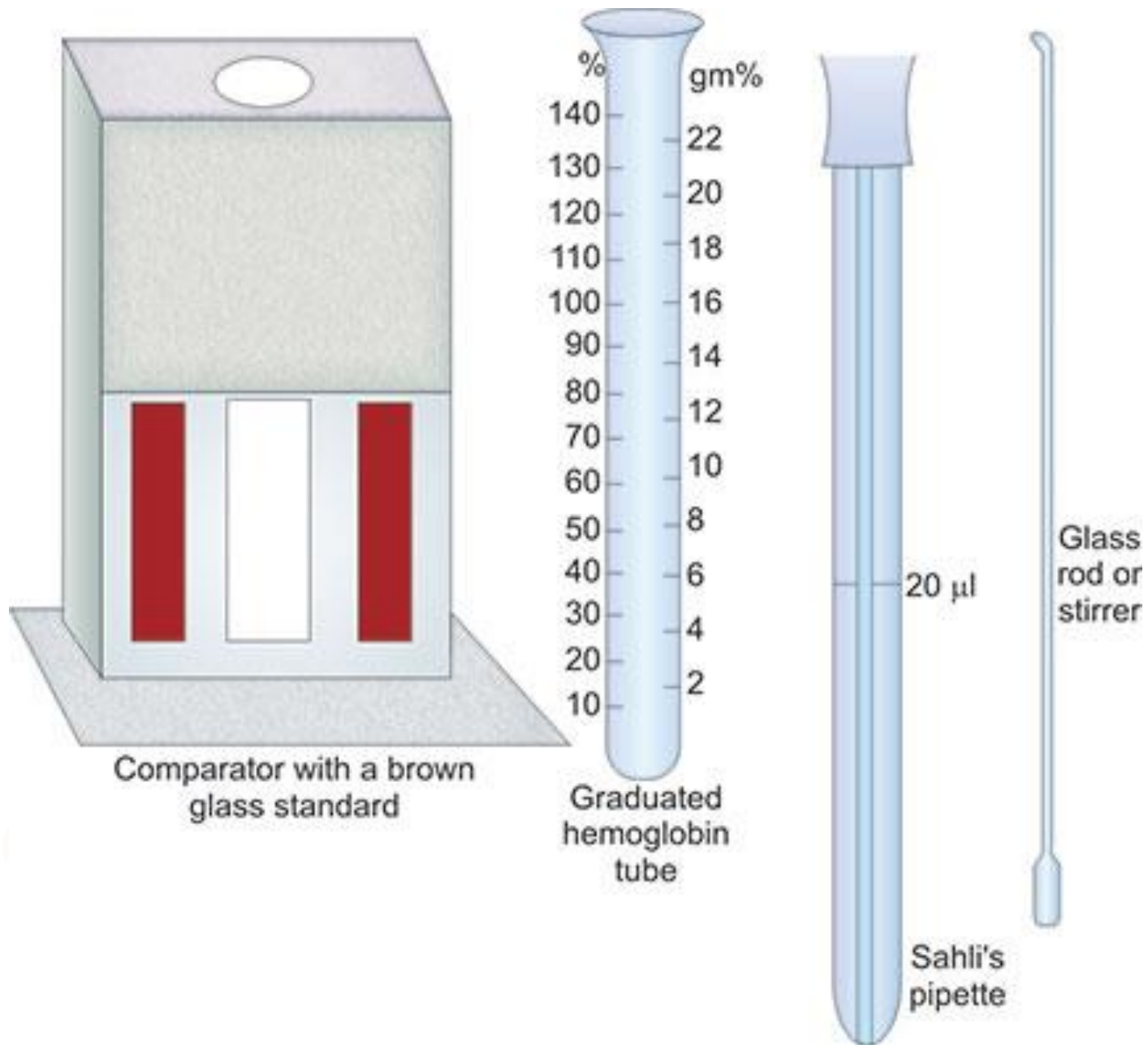
A rare bone marrow disease that leads to an abnormal increase in the number of blood cells (polycythemia vera)

The body having too little water and fluids (dehydration)

high and physical conditions

# Sahli's Acid Hematin Method

- Equipments-
  - Sahli hemoglobinometer
  - Sahli pipette (marked at 20 microlite or 0.02 ml)
  - Stirrer
  - Dropping pipette
- Reagents
  - N/10 hydrochloric acid
  - Distilled water



Sahli's Hemoglobinometer



SUPERIOR

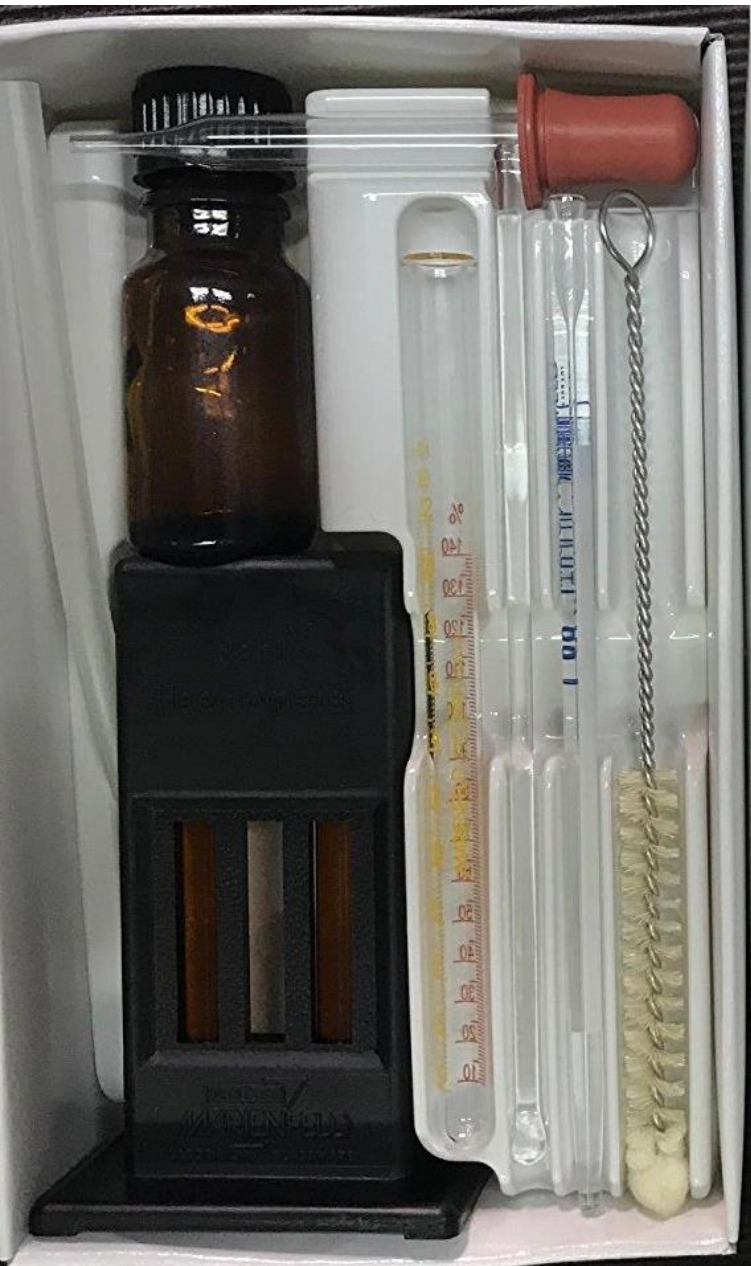
MARIENFELD

Haemometer

**Haemometer acc. to Sahli,** complete set consisting of:  
polystyrene support with 2 colored rods and opal glass  
plate, comparator tube, haemoglobin pipette 20 µl with  
an approx. 16 cm long silicone tube and white mouthpiece,  
dropping pipette with rubber teat, stirring rod, acid vial,  
cleaning brush and directions of use, without lancets

Made in Germany

32 430 00



**Principle:**

Haemoglobin (Hb) is converted to acid haematin by addition of 0.1 N hydrochloric acid and resulting brown colour is compared with standard brown glass reference blocks of a Sahli's haemoglobinmeter.

**Method of Use :**

- By using a Pasteur pipette ,add 0.1N HCl in the graduated tube to the mark 10.
- Draw blood upto the 20  $\mu$ l mark in the Sahli Hb pipette and add it to the acid in the tube. Rinse the pipette well, mix the reaction mixture and allow the tube to stand for atleast 10 min.
- Dilute the solution with distilled water by adding few drops at a time until the colour matches with the standard glass reference blocks.
- The matching should be done only against natural light. The level of the fluid is noted at its lower meniscus and reading corresponding to this level on the scale is recorded in g/dl



Thank You